

Techniques for Assessing Teratogenic Effects: Epidemiology

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Epidemiologic studies of malformations can aid in the understanding of human teratogenesis. Employing a variety of approaches epidemiology can develop or test hypotheses concerning possible causes or through surveillance provide data useful for a variety of purposes. Drawing heavily upon our experiences at the Center for Disease Control, this paper reviews some concepts and uses of epidemiology in studies of human teratogenesis.

Epidemiology is the study of the distribution and determinants of disease among populations. It is the study of the causes and manifestations of disease and of their associations. This paper focuses on the use of epidemiology to study human structural malformations and their environmental determinants. Epidemiologic studies of malformations may be classified as descriptive or analytic. In descriptive studies, malformation cases are characterized according to frequency, race, sex, or other variables related to their occurrence; in analytic studies their associations with possible environmental determinants are defined and tested. Another part of the epidemiology of malformations is surveillance, the routine collection, analysis, and reporting of data.

Some concepts should be kept in mind when considering epidemiologic studies of malformations and teratogenesis. First, structural malformations are only one of several pregnancy outcomes influenced by environmental determinants. Others include spontaneous abortion, fetal and infant death, low birth weight, mental retardation, deafness, and blindness. Limiting at-

tention to malformations only excludes other effects of the environment on the fetus. For practical reasons, studies are generally restricted to one outcome, but the other disorders should be kept in mind, particularly when the study design allows consideration of them.

Second, unless there is evidence for grouping malformations, each should be considered separately, even those involving the same body system. Patients with anencephaly-spina bifida (ASB), for example, have a lower proportion of affected males, greater racial variation, and more familial recurrence than patients with hydrocephalus alone (1-4). Similarly, patients with cleft lip with or without cleft palate differ epidemiologically and, therefore, probably etiologically from those with cleft palate only (5). In fact, patients with cleft lip and palate plus other malformations differ epidemiologically from those with facial clefts only (6). Despite the frequent need for larger numbers of cases to study, the cases should not be combined without regard for their possible epidemiologic or etiologic differences.

Third, one malformation can have a variety of causes, and one cause can result in a variety of malformations. Rubella infection during pregnancy, for instance, can cause congenital heart defects, cataracts, mental retardation, and deafness in affected infants. Conversely, congenital cataracts, while caused by the rubella virus, can also

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occur with over 20 other conditions, including Down's syndrome, a chromosomally determined condition, and galactosemia, a genetically determined biochemical abnormality (7).

Last, many factors and teratogenic mechanisms determine the number and type of human malformations caused by exposure *in utero* to an environmental teratogen. Discussion of these factors and mechanisms is beyond the scope of this presentation, but they have been considered in some detail by Wilson (8). Knowledge of these mechanisms must be brought to bear on epidemiologic studies. For instance, an important determinant of malformations is the gestational age at which the fetus is exposed to the teratogen. The now classic example is thalidomide, which produced a variety of structural abnormalities among infants exposed *in utero*. Most typical were reduction deformities of the limbs and anotia; other anomalies included duodenal stenosis and atresia, heart anomalies, facial hemangiomata, and anophthalmia or microphthalmia (9, 10). Early in the studies of thalidomide, investigators reported that at least 139 pregnant women had taken the drug without its causing fetal malformations. Upon establishing more precisely when the mother took it, however, they defined the embryosensitive period as 34-50 days after the first day of the last menstrual period (11). Embryos exposed during this period were malformed; those exposed outside this sensitive period were unaffected. In this instance, the fact of exposure had to be further qualified by the time of exposure. When this was done, the findings were consistent with knowledge about the timing of normal embryogenesis.

Descriptive Epidemiology

Descriptive epidemiology characterizes patients with malformations according to such variables as geographical differences, seasonal patterns, socioeconomic and racial influences, and maternal age effects. Such studies may identify high or low incidence populations which can suggest areas for further work. The vast majority of epidemiologic studies in humans to date have been descriptive.

ASB has been well described epidemiologically because two circumstances have made these defects especially well suited for study: affected infants are easily recognized and reliably reported and cases occur with sufficient frequency that adequate numbers can be assembled for study.

From these descriptive studies have come an epidemiologic picture of ASB which suggests in part an environmental etiology. Pronounced geographic differences exist between countries and even within the same country. In Ireland, for instance, over 1% of newborns are affected with ASB, while in Great Britain a two- to threefold increase in rates occurs in a progression from southeast to northwest (3, 12). In the United States, the ASB incidence declines from east to west with the rates of 1.0 per thousand in the South being twice that in the West (13). Other findings also suggestive of environmental factors include an inverse relationship between ASB incidence and socioeconomic status (14, 15) and incidence changes of epidemic proportions over time (16, 17).

Descriptive studies of malformations in the United States have generally drawn upon three data sources: vital records (birth, death, and fetal death certificates), available hospital records, and special clinical surveys often employing standard infant examinations. Each source has advantages and disadvantages in terms of reporting completeness, uniformity and reliability of diagnosis, and ease with which the data are obtained. Vital records yield rates of 1% to 1.5% for total malformed infants, hospital records yield rates of 2.5% (18-20), and intensive clinical surveys yield rates ranging from 5% to 15% (21-23). These ranges reflect the composite effect of several variables, including the completeness of recognition and recording of malformations, the infant's age at diagnosis, and the definitions used to distinguish malformations, variants, and states of normalcy. The intensive clinical studies provide the highest rates because they insure that every newborn receives a standard examination with special attention to such relatively inconspicuous conditions as ear shape, the configuration of fingers or toes, and the existence of more trivial birth marks. Of the other two sources, vital records are the most deficient (20), yet they are still quite suitable for some uses. For example, studies of seasonality or the effects of maternal age and parity upon the occurrence of some malformations can be done with vital record data.

Another determinant of malformation case rates is the age at which malformations are recognized and still reported (21, 24, 25). Internal anomalies of the cardiovascular, renal, and gastrointestinal systems in particular may not be evident until some weeks, months, or even years after birth. Therefore, rates will be higher from

data sources which provide for ascertainment of cases into later life.

Descriptive studies require sizeable case numbers and knowledge of the characteristics of the general population from which the cases are drawn. For this reason, they often use data available from the Bureau of the Census or from state vital records departments. These populations generally coincide with county, multicounty, or state boundaries and hence are geographically (or population) based.

The use of a geographic base can pose problems of case ascertainment. Identifying and registering cases over a large geographic area requires the cooperation of many hospitals and sometimes other facilities where malformed infants are seen. The involvement of a number and variety of data sources precludes an intensive clinical examination of each infant. Therefore, one must develop some mechanism for obtaining case reports which combines hospital and vital record reporting, realizing that there will be some loss in reporting completeness.

One example of such a community-based surveillance program is the Metropolitan Atlanta Congenital Defects Program (26). The Program is conducted jointly by the Center for Disease Control (CDC), Emory University School of Medicine, and the Georgia Mental Health Institute and covers a five-county area with approximately 25,000 births annually. Infants with malformations are identified by staff members who regularly visit each of the 21 Atlanta area hospitals with obstetrical or pediatric services. The staff register all infants with structural, chromosomal, or biochemical abnormalities diagnosed up to age 1 year. The most completely reported abnormalities are those evident in the newborn period. Data from these case-finding efforts are supplemented by vital records information furnished by the Georgia Department of Human Resources and by laboratory records of chromosomal abnormalities.

Analytic Epidemiology

From descriptive studies, laboratory investigations, clinical reports and other sources, then, come leads about possible human teratogens. These leads generally result in hypothesis testing with analytic studies. The data available from the Atlanta program, for example, have been used for analytic studies of drugs and other environmental teratogens. In 1973, on the heels of a reported association be-

tween parental use of spray adhesive compounds, chromosomal damage, and congenital malformations, we compared time trends for the rate of malformations with the sales of the suspected teratogen (Fig. 1) (27). In the face of a marked increase in sales no change occurred in the rates of all malformations, infants with multiple defects, or any single malformation. To determine the extent to which pregnant women might have been exposed to the compounds, 173 postpartum women in five Atlanta hospitals were interviewed about their exposure to spray adhesive compounds: nine (4.6%) reported having used them at some time. While the lack of change in incidence is evidence against substantial teratogenesis, a small effect could have been present and not detected by this method. However, by using these same data, an estimate also can be made of the maximum teratogenic risks which the compounds pose. In this case, the proportion of women at teratogenic risk was the proportion of women exposed to spray adhesives who had malformed babies. The rarer the defect, the smaller the risk that might be detected. From the interview data it is estimated that some 5% (1400) of the 28,000 Atlanta women delivering babies in 1973 were potentially exposed. If these compounds affected, for example, 2% of the 1400 pregnancies, there would be 28 "extra" cases from exposure. This would appear as a twofold increase over the usual malformation rate of 1 per 1000 or less. (Roughly 95% of the 130 different malformations coded in Atlanta occur at this frequency or less.) In the unlikely event that spray adhesive compounds caused an increase in all malformations, a 10% risk would be detectable. In this instance there would be an added 140 cases (10% of 1400) to the 700 which occur annually. For more intermediate case rates, the teratogenic risks would be between 2% and 10%.

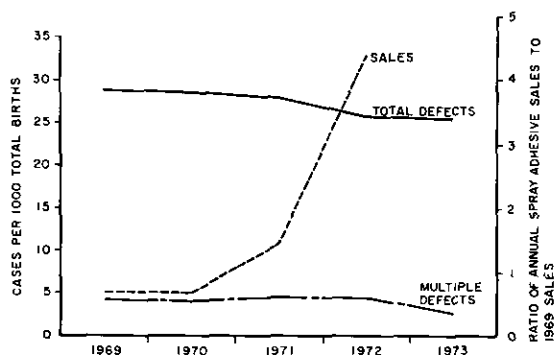


FIGURE 1. Trends in birth defect incidence and spray adhesive sales, metropolitan Atlanta, 1969-1973.

Two other types of analytic studies are the case-control and cohort studies. The case-control study examines a group of cases with 1 or more of the same malformation(s) and a group of suitable controls, comparing their frequencies of exposure to an environmental agent or the presence of some other determinant. Greater frequency of exposure among the case group may suggest a causal association with the malformation under study. Since the data on exposure are collected after the malformation has been diagnosed, the case-control study is in the temporal sense *retrospective*.

Another phase of the Metropolitan Atlanta Congenital Defects Program is the regular interview of mothers 3 months after the birth of an infant with any of 12 selected malformations. Mothers are questioned about their occupation before and during pregnancy, their residence at conception, any illnesses, and any drugs taken during pregnancy. When the interviews began in 1970, Gal et al., also using a case-control study, had reported an association between the use of hormonal tests for pregnancy and the later birth of infants with defects of the central nervous system (28). As part of the Atlanta interview we included a question about the hormonal pregnancy test. Over a 3-yr period, 123 mothers of infants with ASB and 310 mothers of infants with other malformations, who served as controls, were interviewed (Table 1). We chose these mothers as controls because mothers of malformed infants have been shown to recall more events during pregnancy than mothers of normal infants (29). We found no significant differences in frequency of use of the hormonal pregnancy test (30).

The second type of analytic study is the cohort approach. These studies begin with the fact of exposure, for example, a group of pregnant women exposed to an environmental teratogen. These women and a comparison group, preferably similar in all respects except exposure, are then followed prospectively from exposure to outcome. Alternatively, the exposed and unexposed cohorts are analyzed retrospectively after pregnancy has ended. So long as the presence or absence of exposure is defined independent of any knowledge of pregnancy outcome, the "retrospective" nature of such a study need not be reason for skepticism. In fact, this latter approach can be preferable since there need not be provisions for long-term observation of the exposed and non-exposed cohorts.

One notable advantage of cohort studies of teratogenesis is that a variety of poor pregnancy

Table 1. Frequency of use of the hormonal-pregnancy test among mothers of malformed infants by type of malformation (metropolitan Atlanta, Georgia, 1970-73)

Type of malformation	No. of responses	Positive	
		No.	%
Total infants	433	46	10.6
Total neural-tube defects	123	10	8.1
Anencephaly alone	36	2	5.6
Anencephaly and spina bifida	5	0	0.0
Spina bifida alone	78	8	10.3
Encephalocele	4	0	0.0
Cleft lip with or without cleft palate	71	8	11.3
Cleft palate alone	38	5	13.2
Down's syndrome	62	6	9.7
Other chromosomal abnormalities	10	0	0.0
Esophageal atresia	11	3	27.3
Intestinal atresia	38	4	10.5
Omphalocele	34	4	11.8
Diaphragmatic hernia	10	1	10.0
Limb-reduction deformities	20	3	15.0
Syndromes (excluding Down's)	13	1	7.7
Infants with multiple malformations	70	11	15.7

outcomes can be sought among the exposed population. Providing the exposed women are followed from early in pregnancy, the range of effects upon pregnancy, including abortion, stillborn infants, and malformations can be determined. Continued follow-up of infants exposed *in utero* might determine further effects such as mental retardation, learning disabilities, deafness, or blindness. The commonly overriding disadvantage of cohort studies is their expense. The low frequency of most of these conditions necessitates the study of large numbers of persons. Acceptable statistical boundaries for such a study might be a 95% certainty of detecting an effect when present and a 5% risk that an observed effect is falsely positive. Within these limits, a cohort of approximately 700 exposed pregnant women would be required to detect a 10-fold increase in a malformation which occurs normally at a rate of 1 per 1000. Within the same boundaries, detection of a twofold increase would require enrollment of approximately 17,000 exposed women.

Surveillance

Surveillance is a third component of the epidemiology of congenital malformations. Its purposes include the provision of data for

descriptive studies, the development of a registry for case-control studies, and the monitoring of malformation cases for changes suggestive of environmental influences (31). Monitoring and surveillance can be used interchangeably so long as the term conveys a sense of immediacy in the collection and analysis of data. The importance of monitoring has been considered to be the early detection of thalidomide-like epidemics. To date no such episodes have occurred, to our knowledge, so the usefulness of this approach has not yet been tested. Nevertheless, in the face of the countless new substances which current technology contributes to the environment and whose teratogenic effects upon humans are not known, malformation monitoring must be considered prudent for the foreseeable future.

While monitoring of malformation cases can detect changes which might suggest environmental causes, it will not necessarily explain the cause of the change. Vital to the monitoring activity is the systematic comparison of the current incidence of malformations with an expected value based on previous experience. Comparisons should provide for recognition of sudden increases, gradually rising trends, and geographic differences. Because of these many and varied comparisons computers have come to play an important role in monitoring.

The Metropolitan Atlanta Congenital Defects Program has provided for the local monitoring of malformation cases at monthly intervals since 1970. Although many increases have been noted, most were transient, and to date no environmental cause for any of them has been found. Four other programs, in Nebraska, Washington, Upper New York State, and northern Florida, have also contributed to the monitoring effort in this country. The Nebraska and Florida programs use hospital records, while the New York and Washington programs rely upon vital records (31).

The first nationwide monitoring effort was started at the Center for Disease Control in December 1974 with the implementation of the Birth Defects Monitoring Program (BDMP) (partially funded by Interagency Agreement No. YO-HD-31002) (13). This program was developed jointly by CDC, the National Institute for Child Health and Human Development, the National Foundation-March of Dimes, and the Commission on Professional and Hospital Activities (CPHA), a health-data-processing organization located in Ann Arbor, Michigan. Data used in the BDMP are

obtained from hospital discharge records of some 1,200 hospitals located throughout the country. These BDMP hospitals are among those already enrolled in CPHA's Professional Activity Study (PAS), a computerized service for participating hospitals.

The BDMP uses data on newborn discharges already sent to CPHA by the PAS hospitals. There are presently some 1 million births annually in BDMP-PAS participating hospitals. Because of the self-selection of these hospitals, the data are not a random sample of U.S. births nor are they geographically based. Nevertheless, the BDMP represents the largest single source of uniformly collected and coded data presently available on malformations among newborns in the United States. The percentage of live births by census division in the BDMP varies from over 15% in the East and West South Central Divisions to 51% in the East North Central Division (Fig. 2). This variation reflects the self-selection of hospitals which choose first and primarily to participate in the PAS and, second, in the BDMP. For a case with a malformation to be used in the BDMP system, the defect must be apparent at birth or during the newborn's nursery stay, be noted by the attending physician, and be recorded by the medical record department staff on discharge abstracts routinely sent to CPHA.

At CPHA a special data file is maintained for all newborns discharged from BDMP hospitals with any of some 200 different defects. Data already available for 1970 through 1973 were entered into this special file and are used as the baseline rate for calculating the expected numbers of malformation cases. At quarterly intervals, for each defect category the most recently observed number of cases is compared with

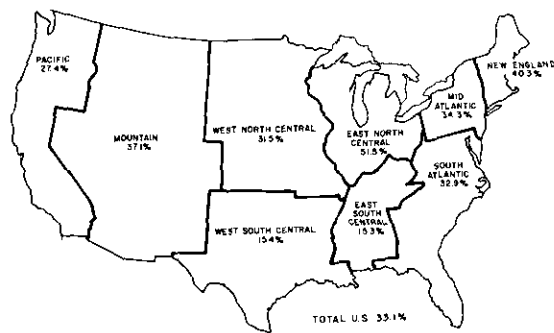


FIGURE 2. Percent of U.S. live births in the Birth Defects Monitoring Program, by U.S. Census Division, January-June 1975.

the expected. Comparisons are made for 3-, 6-, and 12-month time periods and for geographic areas ranging from individual county to the whole United States. Whenever the observed number of cases in any of these exceeds the expected number at the 0.01 level or greater, the defect and the geographic area are listed on an exceptions report by the computer. These lists along with other data are then sent to CDC for analysis and evaluation for possible environmental causes.

One of the more striking increases observed early was a three- to four-fold increase in the incidence of lung agenesis which occurred between 1973 and 1974 rather uniformly throughout the United States (Fig. 3). Upon further inquiry it was noted that a revision of the H-ICDA code (32) instituted on January 1, 1974, had resulted in the assignment of lung hypoplasia, a different but related defect to the lung agenesis category.

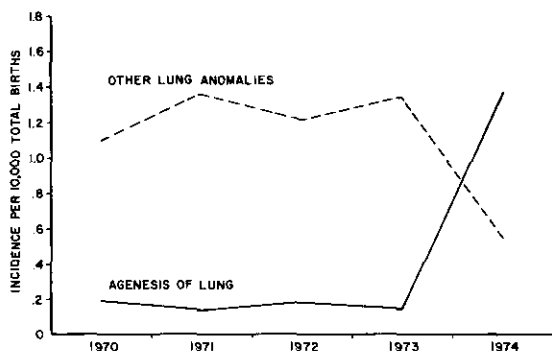


FIGURE 3. Annual incidence of lung agenesis and other lung anomalies, U.S. 1970-1974.

Although in this instance a coding circumstance caused an apparent increase, the episode shows that the BDMP can detect these sudden changes. This example also shows the need to know intimately the strengths, weaknesses, and intricacies of one's data source. A more gradual, but progressive increase in heart anomalies has also been noted, exemplified by a steady rise since 1970 in the rates for ventricular septal defect (Fig. 4). Similar increases have been noted for PDA, tetralogy of Fallot, and total heart anomalies. We have no definite explanation for these increases but believe them to be attributable to better diagnosis and reporting. During this same period there have been more newborn intensive care nurseries established and more aggressive care of premature and other sick infants. Both would presumably result in greater opportunity for detection and follow-up of cases

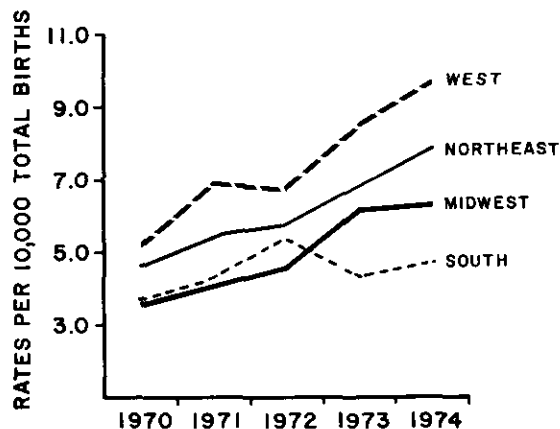


FIGURE 4. Ventricular septal defect incidence rates, by census region, U.S. 1970-1974.

with these conditions. However, a definitive explanation can come only from special clinical studies involving a large number of infants and providing for special follow-up of cases.

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